### **Research** Article



### Biomass Yield Response of Different Medicinal Plants Under Dual Stress of Salinity and Sodicity

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**Abstract** | One of the best strategies for the utilization of salts affected soils is the screening of available local plants which can grow or survive under salt stress and have considerable economic importance to the farming community. Therefore, a three-years pot experiment was executed to explore the salinity tolerance of medicinal plants i.e., Podeena (*Mentha spicata*), Hina (*Lawsonia inermis*), Qulfa (*portulace oleracea*), Methi (*Trigonella foenumgraceum*), Dill (*Anethum graveolens*) and Kalwanji (*nigella sativa*), under dual stress of EC (electrical conductivity of soil extract) 0.79, 6 and 8 dS m<sup>-1</sup> and SAR (sodium adsorption ratio) 5.99, 25 and 35. Each crop was grown for four months and biomass yield data was recorded. Results of three successive seasons suggest that all the evaluated medicinal plants can grow under the medium salinity and sodicity level of (6 dS m<sup>-1</sup> + 25 SAR). However, biomass yield decreased linearly with increasing levels of salinity and sodicity and a maximum reduction of 63.25% for Podeena, 48.15% for Hina, 54.74% for Qulfa, 32.87% for Methi, 59.77% for Dill and 45.18% for Kalwanji was recorded at the highest level of salinity + sodicity (EC 8 dS m<sup>-1</sup> + SAR 35).

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### Introduction

S alt stress is one of the main limiting factors for plant production that limits the spread of plants in dryland regions. It is likely to cause further deterioration in crop growth and production due to uncertain precipitation patterns and a rise in temperature expected from climate change (Panta *et al.*, 2014). According to Wang *et al.* (2003), salinization will affect half of the cultivatable area in the middle of the 21<sup>st</sup> century. At the same time, to ensure food security, agriculture has expanded into marginally salt-affected areas that could support biomass production (Shabala, 2013). One of the best

and most practical approaches to manage the salinity problem is the screening of available local cash crops which can grow or survive in salinized environments. In this perspective, medicinal plants are worthy of attention to become a promising candidate for cultivation in marginally salt-affected areas because of their high economic value and the global trade of medicinal plant that is expected to reach \$ 5 trillion (US) by 2050 (Shinwari, 2010). In 2012, Pakistan exported medicinal plants costing over the US \$10.5 million (MINFAL, 2012). Therefore, knowledge of the salinity tolerance potential of traditionally used medicinal plants is necessary for the economic utilization of saline areas and the betterment of socioeconomic conditions of the farming community.

In a pot experiment, Hussain et al. (2009) studied the growth performance of black cumin (Nigella sativa L.) under salinity stress of 0, 3 and 6 dS m<sup>-1</sup>. They reported that salinity had adverse effects on growth attributes, and maximum reduction in shoot length and fresh and dry weight of black cumin was at salinity stress of 6 dS m<sup>-1</sup>. Likewise, Moosavi et al. (2013) assessed the effect of salt stress (0, 2, 4, 6 and8 ds m<sup>-1</sup>) on germination and growth parameters of black cumin. They concluded that the highest level of salinity (8 ds m<sup>-1</sup>) reduced seedling weight (33.3%), seedling length (21%) and germination percentage (13.2%) over the control. Similarly, Faravani et al. (2013) stated that salt stress of 15 dS m<sup>-1</sup> caused a significant negative effect on biomass yield, plant height and biological yield of black cumin.

In a greenhouse study, Roodbari et al. (2013) irrigated the mint plants with 0, 50, 100 and 200 mmol NaCl solution. Increasing levels of NaCl salinity caused a remarkable reduction in root and shoot growth. In addition, peppermint did not survive at 200 mmol NaCl. Shayghan and Sedghi (2013) evaluated the performance of mint against five salinity levels (0, 2, 4, 6 and 8 ds m<sup>-1</sup>). They reported that the highest level of 8 dS m<sup>-1</sup> prompted adverse effects on seed viability index, germination rate and germination percentage of mint. Purslane as compared to any other vegetable is accepted as a comparatively more salt-tolerant medicinal plant (Kiliç et al., 2010). The growth performance of 25 purslane accessions under NaCl stress (0, 10, 20, 30 and 40 dS m<sup>-1</sup>) was evaluated by Alam et al. (2014). Plant height, dry matter yield, the number of flowers and the number of leaves decreased significantly with increasing salinity stress.

They opined that Ac7 and Ac9 were comparatively more salt tolerant among all 25 purslane accessions.

Ratnakara and Rai (2013) studied the effects of different levels of salt stress (0 mM to 100 mM NaCl) on the early growth stages of Triginella foenum. Results showed that 40 mM NaCl did not affect germination, however, at a higher level of 80 mM NaCl, seeds were unable to germinate. A gradual decrease in biomass yield of Triginella foenum was observed with increasing levels of salinity. Garg (2012) investigated the effect of sodicity (10, 20, 30, 40 and 50) exchangeable sodium percentage (ESP) on four varieties of fenugreek (Kalyanpur Selection, RMt-1, Hissar Sonali and HM-346). The yield parameters were unaffected up to ESP 30, however, increasing levels of ESP significantly reduced the yield attributes of all varieties. Kalyanpur Selection recorded the maximum biomass and seed yield than the other varieties.

Keeping in view the importance of medicinal plants as cash crops, the study was planned to assess the effect of different salinity/ sodicity levels on the biomass yield of tested medicinal plants and to identify the level at which these medicinal plants can grow successfully in saline-sodic conditions.

### **Materials and Methods**

A pot experiment was conducted in the wire house of Soil Salinity Research Institute, Pindi Bhattian, Pakistan (latitude 31.8950° N and longitude 73.2706° E), for three consecutive seasons. A normal soil was collected, air dried, passed through a 2 mm sieve and analyzed by following the methods of U.S. Salinity Laboratory Staff (1954), properties are given in Table 1. Soil pH of the saturated paste was measured by using pH meter (Microcomputer pH-vision cole parmer model 05669-20). Electrical conductivity of the irrigation water and soil saturated paste extract was measured with the help of conductivity meter (WTW conduktometer LF 191). The Na<sup>+</sup> contents were determined by flame photometer (digiflame code DV 710) while Ca2+ and Mg2+ were determined titrimetrically. Sodium adsorption ratio (SAR) was calculated as follows. SAR = Na<sup>+</sup> /  $[(Ca^{2+} Mg^{2+})^2]^{1/2}$ .

Desired levels of  $EC_{e}$  and SAR were developed artificially for each medicinal crop in each season for three years by using Na<sub>2</sub>SO<sub>4</sub>, NaCl, CaCl<sub>2</sub> and MgSO<sub>4</sub> as calculated with the help of quadratic equation (Ghafoor et al., 1988). Treatments included were:  $T_1 = EC_e 0.79 \text{ dS m}^{-1} + \text{SAR } 5.99, T_2 = EC_e 6 \text{ dS m}^{-1} + \text{SAR } 25, T_3 = EC_e 6 \text{ dS m}^{-1} + \text{SAR } 35, T_4^{-1} = EC_e 8 \text{ dS m}^{-1} + \text{SAR } 25, T_5 = EC_e 8 \text{ dS m}^{-1} + \text{SAR } 35.$ 

Table 1: Initial soil analysis at the sta	irt of study.
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Parameter	Value
pH <sub>s</sub>	7.80
EC <sub>e</sub> (dS m <sup>-1</sup> )	0.79
SAR	5.99
Saturation percentage	28.80
Texture	Sandy loam
Organic matter (%)	0.75
Available K (mg kg <sup>-1</sup> )	120.0
Available P (mg kg <sup>-1</sup> )	8.40

After the development of desired levels of salinity and sodicity, glazed pots were filled @ 20 kg soil per pot. In Kharif season Podeena (Mentha spicata), Hina (Lawsonia inermis), and Qulfa (Portulaca oleracea), while in Rabi season Methi (Trigonella foenumgraecum), Dill (Anethum graveolens), and Kalvanji (Nigella sativa) were sown. Ten seeds of each crop were sown in each pot. Completely Randomized Design (CRD) was applied with four replications. Tap water {SAR= 3.39, EC = 0.77 (dS m<sup>-1</sup>) and RSC= 0.85 (me  $L^{-1}$ ) was used to irrigate the pots. Thirty days after sowing, seedlings were thinned and four seedlings per pot were maintained. Crops were grown for four months and biomass yield data were recorded. Collected data were statistically analyzed following analysis of variance (ANOVA) and means were compared by LSD at alpha 0.05 (Steel et al., 1997).

### **Results and Discussion**

#### Podeena

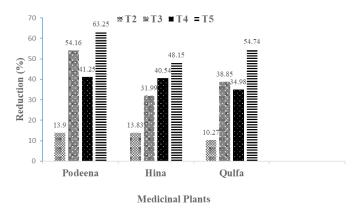
Based on the results of three seasons, data about the biomass of Podeena revealed that salinity-sodicity clearly arrested the growth of Podeena plants (Table 2). The maximum biomass yield (13.09 g) was divulged by control (non-saline) which remained non-significant with  $T_2$ , however, further increased salinity-sodicity stress decreased the biomass yield significantly (p < 0.05). The minimum biomass yield of 4.81 g was documented at the highest salinity-sodicity condition of EC<sub>e</sub> 8 dSm<sup>-1</sup> and SAR 35 ( $T_5$ ). Under the salinity-sodicity conditions, biomass yield

decreased by 13.90%, 54.16%, 41.25% and 63.25% respectively in  $T_{2}$ ,  $T_{3}$ ,  $T_{4}$  and  $T_{5}$  when compared with the control (Figure 1).

### **Table 2:** Effect of salinity / sodicity on Podeena (Mentha spicata) biomass yield $(g \text{ pot}^{-1})$ .

Treatments EC: SAR				Mean	Percent de- crease over control
T <sub>1</sub> -Control	12.42 A	13.56A	13.30A	13.09 A	
$T_2^{-(6:25)}$					
$T_{3}^{-}(6:35)$	5.50CD	6.73BC	5.76B	6.00 BC	54.16
T <sub>4</sub> -(8:25)	7.36BC	8.30B	7.43B	7.69 B	41.25
T <sub>5</sub> -(8:35)	4.43 D	4.96 C	5.03B	4.81 C	63.25

Different letters in each column indicate significant differences among the treatments at  $p \le 0.05$ .



**Figure 1:** Percent reduction (over control) in biomass yield of Podeena, Hina and Qulfa with increasing levels of EC<sub>e</sub> and SAR (average of three seasons).  $T_2$  (EC<sub>e</sub> 6 (dSm<sup>-1</sup>) + SAR 25),  $T_3$  (EC<sub>e</sub> 6 (dSm<sup>-1</sup>) + SAR 35),  $T_4$  (EC<sub>e</sub> 8 (dSm<sup>-1</sup>) + SAR 25),  $T_5$  (EC<sub>e</sub> 8 (dSm<sup>-1</sup>) + SAR 35).

### Hina

Saline-sodic stress conditions significantly decreased (p < 0.05) the biomass yield of Hina (Table 3). The mean value of three seasons indicated the maximum biomass yield (9.47 g) was produced by control, which was at par (p < 0.05) with a medium salinity-sodicity level (T<sub>2</sub>). On contrary, biomass yield decreased significantly with the higher levels of EC<sub>e</sub> and SAR while minimum biomass yield (4.91 g) was produced by T<sub>5</sub> with (EC<sub>e</sub> 8 dS m<sup>-1</sup> and SAR 35). Yield reduction of 13.83%, 31.99%, 40.54% and 48.15% was observed in T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub> and T<sub>5</sub> when compared with non-stressed plants (control) (Figure 1).

### Qulfa

Biomass yield of Qulfa was also affected adversely by the dual stress of salinity-sodicity and the negative effect was more pronounced at the highest levels of EC<sub>e</sub> and SAR. Pooled data of three seasons (Table 4) indicated that non stress plants recorded the maximum biomass yield of 10.32 g with no difference (p < 0.05) from medium salinity sodicity levels (T<sub>2</sub>). While the highest levels of EC<sub>e</sub> 8 dS m<sup>-1</sup> and SAR 35 recorded a minimum yield of 4.67 g. Salinity-sodicity led to a reduction of 10.27%, 38.85%, 34.98% and 54.74%, respectively in T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub> and T<sub>5</sub> in comparison to control plants (Figure 1).

# **Table 3:** Effect of salinity / sodicity on Hina (Lawsonia inermis) biomass yield (g $pot^{-1}$ ).

Treatments EC: SAR				Mean	Percent decrease over control
T <sub>1</sub> -Control	9.02 A	10.13 A	9.26 A	9.47 A	
$T_2$ -(6:25)	5.16 B	10.10 A	9.23 A	8.16 A	13.83
$T_3 - (6:35)$	4.47 B	7.50 B	7.36 AB	6.44 B	31.99
T <sub>4</sub> -(8:25)	5.08 B	5.80 BC	6.03 BC	5.63BC	40.54

Different letters in each column indicate significant differences among the treatments at  $p \le 0.05$ .

### **Table 4:** Effect of salinity/ sodicity on Qulfa (Portulace oleracea) biomass yield ( $g \text{ pot}^{-1}$ ).

Treatments EC: SAR					Percent decrease over control
T <sub>1</sub> -Control	10.37A	9.63 A	10.96A	10.32A	
$T_2^{-}(6:25)$	9.98 A	8.20 B	9.60 A	9.26 A	10.27
$T_{3}$ -(6:35)	6.50 B	6.06 C	6.36BC	6.31 B	38.85
T <sub>4</sub> -(8: 25)	6.67 B	7.03BC	6.43 B	6.71 B	34.98
T <sub>5</sub> -(8:35)	4.80 C	4.63 D	4.60 C	4.67 C	54.74

Different letters in each column indicate significant differences among the treatments at  $p \le 0.05$ .

# **Table 5:** Effect of salinity / sodicity on Methi (Trigonella foenumgraceum) biomass yield ( $g \text{ pot}^{-1}$ ).

Treatments EC: SAR				Mean	Percent decrease over control
$T_1$ -Control	26.80 A	21.47 A	23.10 A	23.79A	
$T_2^{-}(6:25)$	22.48 B	18.62 B	21.10AB	20.73B	12.86
$T_{3}$ -(6:35)	19.00 C	15.23CD	15.66 C	16.63C	30.09
T <sub>4</sub> -(8:25)	20.03BC	16.72 C	17.06BC	17.94C	24.59
T <sub>5</sub> -(8:35)	17.71 C	13.90 D	16.30 C	15.97C	32.87

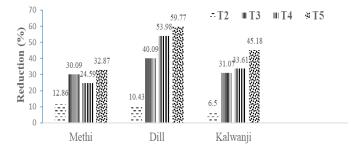
Different letters in each column indicate significant differences among the treatments at  $p \le 0.05$ .

#### Methi

Mean value data in Table 5 exhibited that combined

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stress of salinity and sodicity reduced the biomass yield of Methi and reduction was more remarkable in plants subjected to the highest level of EC<sub>e</sub> and SAR. The non-saline condition produced the maximum (23.79 g) biomass yield, which decreased linearly with increasing levels of salinity-sodicity and minimum biomass yield (15.97 g) was produced at EC<sub>e</sub> 8 dS m<sup>-1</sup> and SAR 35 in T<sub>5</sub>. As compared to control, a reduction of 12.86%, 30.09%, 24.59% and 32.87% was noted in T<sub>2</sub> T<sub>3</sub>, T<sub>4</sub> and T<sub>5</sub> (Figure 2).



**Medicinal Plants** 

**Figure 2:** Percent reduction (over control) in biomass yield of Methi, Dill and Kalwanji with increasing levels of EC<sub>e</sub> and SAR (average of three seasons).  $T_2$  (EC<sub>e</sub> 6 ( $dSm^{-1}$ ) + SAR 25),  $T_3$  (EC<sub>e</sub> 6 ( $dSm^{-1}$ ) + SAR 35),  $T_4$  (EC<sub>e</sub> 8 ( $dSm^{-1}$ ) + SAR 25),  $T_5$  (EC<sub>e</sub> 8 ( $dSm^{-1}$ ) + SAR 35).

**Table 6:** Effect of salinity / sodicity on Dill (Anethum graveolens) biomass yield  $(g \text{ pot}^{-1})$ .

Treatments EC: SAR				Mean	Percent decrease over control
T <sub>1</sub> -Control	23.67A	25.83A	23.53A	24.34A	
$T_2^{-}(6:25)$	17.26B	24.43A	23.73A	21.80A	10.43
$T_{3}^{-}(6:35)$	14.32C	14.66B	14.76B	14.58B	40.09
T <sub>4</sub> -(8:25)	10.56D	10.96BC	12.10BC	11.20C	53.98
T <sub>5</sub> -(8:35)	9.87D	10.03C	9.46C	9.79C	59.77

Different letters in each column indicate significant differences among the treatments at  $p \le 0.05$ .

### Dill

Data about biomass yield of dill plants (Table 6) displayed that the growth performance of Dill plants was very good under normal soil conditions (non-salinized). On the other hand, salinity-sodicity had remarkedly decreased the biomass yield of Dill plants. The maximum biomass yield of 24.34 g was recorded by control plants ( $T_1$ ), whereas minimum biomass yield (9.79 g) was observed at the highest intensities of salinity-sodicity ( $T_5$ ). Biomass yield of Dill was reduced by 10.43%, 40.09%, 53.98% and 59.77% respectively in  $T_2$ ,  $T_3$ ,  $T_4$  and  $T_5$  as compared to  $T_1$ 





### Kalwanji

A negative impact of salinity-sodicity stress was also observed on the biomass yield of Kalwanji (Table 7). Control plants ( $T_1$ ) recorded the maximum biomass yield (15.38 g) with no difference to  $T_2$  (EC<sub>e</sub> 6 dS m<sup>-1</sup> and SAR 25). Contrary, plant subjected to salinitysodicity stress of EC<sub>e</sub> 8 dS m<sup>-1</sup> and SAR 35, produced the minimum biomass yield (8.43 g). Salinity-sodicity stress reduces the biomass yield of Kalwanji by 6.50%, 31.07%, 33.61% and 45.18% respectively in  $T_2$ ,  $T_3$ ,  $T_4$ and  $T_5$  when compared with control (Figure 2).

## **Table 7:** Effect of salinity / sodicity on Kalwanji (Nigella sativa) biomass yield (g pot<sup>-1</sup>).

Treatments EC: SAR				Mean	Percent decrease over control
$T_1$ -Control	15.03A	16.50A	14.63A	15.38A	
$T_2^{-}(6:25)$	13.43B	15.23A	14.50A	14.38A	6.50
$T_3$ -(6:35)	10.87C	11.26B	9.66B	10.60B	31.07
T <sub>4</sub> -(8:25)	10.51C	10.13BC	10.00B	10.21B	33.61
T <sub>5</sub> -(8:35)	8.47D	8.43C	8.40B	8.43C	45.18

Different letters in each column indicate significant differences among the treatments at  $p \le 0.05$ .

The increasing demand for medicinal herbs in the pharmaceutical industry has led to an increase in their cultivation area. Therefore, the study of their salinity tolerance can be a strategic approach for the profitable use of salt-affected soils rather than neglecting this valuable natural resource. The tested herbal plants are well known for their medicinal properties. So, knowledge of their salt tolerant potential may offer the opportunity of an alternative and promising cash crop for marginally salt-affected soils. In this study, biomass production of medicinal plants was considered an aspect of salt tolerance. Hence, the performance of six medicinal plants in term of biomass production was evaluated against five different combinations of salinity and sodicity. In current study, no significant difference in biomass yield of tested medicinal plants was observed at (6 dS m<sup>-1</sup> + 25 SAR) in comparison to control (nonsalinized). However, a negative correlation between biomass yield and the increasing levels of salinity and sodicity was found whereas, maximum reductions of 63.25% for Podeena, 48.15% for Hina, 54.74% for Qulfa, 32.87% for Methi, 59.77% for Dill and 45.18% for Kalwanji was evident at EC 8 dS m<sup>-1</sup> +

SAR 35. Earlier, Saberali and Moradi (2019) found that Dragonhead, Dill, Fenugreek and Savory were moderately tolerant to 40 mM NaCl, while further increase from 40 to 160 mM NaCl diminished the seedling mass from 5 to 63% for the Fenugreek, 4 to 67% for the Dill, 10 to 31% for the Dragonhead and 12 to 71% for the Savory.

This reduction in biomass yield of all the tested medicinal plants could be explained owing to inability of roots to uptake water and essential plant nutrients (Munns and Tester, 2008) due to the saltinducing osmotic effect, a phenomenon caused by salt stress. A reduced osmotic potential is caused by excessive salts in the growing media. Shoot growth is the result of cell division and enlargement while excess of salts in rhizosphere reduces the turgor pressure, inhibit cell division and growth and the suite of metabolic processes (Munns, 2002; Morais et al., 2012). Consequently, less carbon is available for growth and extra energy is needed by the plant cell (Razmjoo, 2008; Kelepesi and Tzortzakis, 2009). In addition, reduced water uptake induced the stomata closure, reduced the uptake of CO<sub>2</sub> and thus disrupting the photosynthetic activity needed for plant growth (Greenway, 1980). Reports claiming that salt stress depressed the growth of medicinal plants are available for Dill (Zehtab-Salmasi, 2008), Thyme species (Belaqziz et al., 2009), Chamomilla recutita L (Ghanavati and Sengul, 2010), Aloe vera (Moghbeli et al., 2012) Fenugreek (Ratnakar and Raib, 2013), Citronella java (Chauhan and Kumar, 2014) and Cumin (Hassanzadehdelouei et al., 2013).

In addition, secondary effects of salinity may arise, involving uptake of toxic salts which negatively affect the plant performance (Acosta-Motos *et al.*, 2017). Excessive uptake of Na caused nutritional imbalance and detriment K uptake, increased reactive oxygen species, damage the membrane structure and reduced chlorophyll content which may affect the plant growth and survival (Gerona *et al.*, 2019; Hniličková *et al.*, 2019). Chrysargyris *et al.* (2021) opined that salinity level of 75 mM NaCl decreased the biomass yield of verbena and geranium by 38.2% and 21.5%, respectively. Salt stress of 4.5 g/L of NaCl decreased the plant height of apple mint and pennyroyal by 50% (Aziz *et al.*, 2008).

Plants exposed to salinized environment exhibit the nutritional disorder and availability of essential plant

nutrient like N, P, Ca and Mg decreased (Dorais et al., 2001) which may result the poor growth of plants. In salt stressed plants reduced photosynthetic activity is another critical aspect responsible for reduced plant growth and productivity (Zhao et al., 2007). Excessive accumulation of Na disturbs the biosynthesis of chlorophyll content, lowered the stomatal conductance and net photosynthetic rate (Chrysargyris et al., 2021). When accumulated Na or Cl rise to toxic levels in leaf tissue, it initiates necrotic tips or margins or premature leaf senescence (Hniličková et al., 2019), thus leading to stunted growth. Similarly, Tabatabaie et al. (2007) detected a remarkable reduction in fresh weight of peppermint grown at 2.8 and 5.6 ds m<sup>-1</sup>. Growth characteristics (capitula and leaves per plant, plant height and stem diameter) of milk thistle were reported to be repressed when exposed at salinity of 9 ds m<sup>-1</sup>(Ghavami and Ramin, 2008).

### **Conclusions and Recommendations**

It is imperative to determine safe limit of the environmental stresses like salinity at which medicinal plants give higher yields with better quality. Results of our study suggested that all the evaluated medicinal plants can grow under the medium salinity cum sodicity level of 6 dS m<sup>-1</sup> + 25 SAR. However, salinity and sodicity level of 8 dS m<sup>-1</sup> + 35 SAR reduces the biomass yield of Podeena (63.25%), Hina (48.15%), Qulfa (54.74%), Methi (32.87%), Dill (59.77%) and Kalwanji (45.18%) over control. Further field studies are required to confirm their immense potential to be utilized on salt affected soils as valuable resource and cash crop on an urgent basis.

### **Novelty Statement**

Evaluated medicinal plants can grow under the medium salinity + sodicity level of 6 dS  $m^{-1}$  + 25 SAR without significant loss in biomass yield.

### Author's Contribution

GQ and KA: Conducted the study and original draft. MAQ, HR, MZM, AIS: Reviewed the article. MQN, MR, MFN, and QMA: Collected the data. MAA, AW and GS: Provided the technical input.

### Conflict of interest

The authors have declared no conflict of interest.

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